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Cascading Indirect Effects in a Coffee Agroecosystem: Effects of Parasitic Phorid Flies on Ants and the Coffee Berry Borer in a High-Shade and Low-Shade Habitat

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ABSTRACT Nonconsumptive effects (NCE) of parasites on hosts vary with habitat complexity thereby modifying trait-mediated effects on lower trophic levels. In coffee agroecosystems, *Pseudacteon* sp. phorid fly parasites negatively affect *Azteca instabilis* F. Smith ants via NCE thereby indirectly benefiting prey. It is unknown how differences in habitat complexity influence *Azteca*-phorid interactions or how phorids affect the coffee berry borer (*Hypothenemus hampei* Ferrari), an important pest of coffee (*Coffea arabica* L.). We tested the following hypotheses in field and lab experiments to find the impact of NCE of phorids on *A. instabilis* and trait-mediated indirect effects of phorids on the coffee berry borer: (1) Phorid effects on *A. instabilis* differ between complex and simple shade habitats and (2) Phorids, by modifying *A. instabilis* behavior, indirectly affect coffee berry borer abilities to invade coffee berries. Phorids had greater impacts on *A. instabilis* activity in low-shade farms, but differences in phorid impacts were not mediated by phorid density or light availability. In the lab, phorids had strong cascading effects on abilities of *A. instabilis* to deter coffee berry borers. Without phorids, *A. instabilis* limited coffee berry borer attacks, whereas when the coffee berry borer was alone or with *A. instabilis* and phorids, more coffee fruits were attacked by coffee berry borer. These results indicate that *A. instabilis* has stronger biological control potential in high-shade farms, but the exact mechanism deserves further attention.

KEY WORDS agroforestry, *Azteca instabilis*, nonconsumptive effects, *Pseudacteon*, trait-mediated interactions

Density and trait-mediated indirect interactions are pervasive in terrestrial and aquatic communities, and frequently result in trophic cascades (Abrams 1995, Werner and Peacor 2003, Schmitz et al. 2004). Density-mediated indirect interactions (DMII) result from predators consuming prey leading to a release of the prey's food (Abrams 1995, Werner and Peacor 2003). Trait-mediated indirect interactions (TMII), however, result from predator-induced changes in prey or host phenotype including developmental, morphological, physiological, and behavioral characteristics (Trussell et al. 2002, Werner and Peacor 2003). Both DMII and TMII may result in species-level trophic cascades where changes in predator numbers influence the success of herbivores and plants (Polis et al. 2000). In the case of trophic cascades resulting from TMII, predators have a nonconsumptive effect (NCE) on prey (e.g., herbivores) resulting in a change at a lower trophic level (e.g., plants). NCE are defined as direct effects of predators on prey accomplished by modifying a prey trait (Abrams 2007). Although DMII and TMII are more commonly discussed in the context of predator-prey interactions, host-parasite interac-

tions may also involve important TMII. For example, in host-parasitoid systems, TMII are evident in apparent competition or when plant volatiles released in response to herbivory attract parasitoids (Müller and Godfray 1999). Phorid flies are important parasites of ants, and impact hosts via 'consumptive' and 'nonconsumptive effects'. *Pseudacteon* phorids generally lay eggs in ant thoraxes that as larvae migrate to the heads; ultimately killing ants (Consoli et al. 2001) but phorid presence near an ant colony also results in a trait-modification or NCE on the ants (Folgarait and Gilbert 1999, Feener 2000, Wuellner et al. 2002, Philpott et al. 2004). Namely, when ants detect phorid presence in the area, ant activity and foraging is limited, thereby allowing herbivores increased access to plants (Philpott et al. 2004).

The environmental context in which predator-prey interactions or parasite-host interactions takes place can determine the strength of NCE and TMII. Direct consumptive effects of predators on prey differ with habitat complexity (e.g., via lower encounter rates, predator-avoidance, prey refuges [e.g., Sih et al. 1998]), but relatively few examples examine how habitat complexity may affect NCE or TMII. In littoral habitats, dragonfly nymphs cause movement of mayfly

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larvae facilitating predation by bluegill sunfish, but only in areas with simple vegetation (Swisher et al. 1998). In more complex habitats, mayflies presumably find refuge from bluegill predators. In contrast, in oyster reefs NCE of toadfish (predators) on oyster predator avoidance behavior did not vary with habitat complexity (Grabowski 2004, Grabowski and Kimbro 2005). Direct parasitic effects of parasitoids may be stronger in less complex habitats (Marino and Landis 1996, but see Menalled et al. 1999). At least one example shows that habitat type influences NCE of parasites on hosts. *Apocephalus* spp. phorid fly parasites of the two ant species *Pheidole diversipilosa* Wheeler and *Pheidole bicarinata* Mayr restricted the foraging and number of soldier ants on baits in simple habitats more than in complex habitats (Wilkinson and Feener 2007). Additionally, phorids attacked ants significantly less often in areas that contained more leaf litter compared with areas with little foliage (Wilkinson and Feener 2007). Thus, the potential exists for variation in NCE of parasites on hosts depending on habitat complexity.

We studied a component of an insect food web that involves undescribed species of parasitic phorid flies, *Pseudacteon* spp., the host, the tree-nesting ant, *Azteca instabilis* F. Smith, and an important coffee (*Coffea arabica* L.) pest in coffee agroecosystems varying in habitat complexity in Chiapas, Mexico. *A. instabilis* is an aggressive ant that may control important coffee pests (Perfecto and Vandermeer 2006). Phorids have strong NCE on *A. instabilis*, limiting ant activity level by about half, which results in indirect positive effects on insects usually attacked by ants, such as lepidopteran larvae and ladybird beetles (Philpott et al. 2004, Liere and Larsen 2010). There is limited information, however, on whether or not phorids have these same indirect effects on coffee pests, such as the coffee berry borer (*Hypothenemus hampei* Ferrari). The coffee berry borer is considered the most important pest of coffee worldwide (Soto-Pinto et al. 2002). Previous evidence shows correlations between increased *A. instabilis* activity and decreased coffee berry borer incidence (Perfecto and Vandermeer 2006), yet, there is no direct evidence that *A. instabilis* limits the coffee berry borer or data showing how phorids may affect *A. instabilis*-coffee berry borer interactions. Furthermore, coffee agroecosystems are prone to management changes that affect habitat complexity (Beer et al. 1998). Coffee farms generally include a shade canopy over coffee shrubs below, but canopy complexity (e.g., diversity and density of shade trees, canopy cover, canopy thickness) varies greatly between farms. As coffee farms become more intensely managed with reduced canopy complexity, biodiversity is lost (Perfecto et al. 1996, Moguel and Toledo 1999, Philpott et al. 2008) and complex ecological interactions, such as those involving ants, phorids, and the coffee berry borer may be altered.

We examined the impacts of coffee habitat complexity on NCE of phorids on ants, and subsequent TMII of phorid flies on coffee berry borer via the modification of ant behavior. We tested the following

two hypotheses: (1) Phorid NCE on *A. instabilis* ants differ between complex and simple shade habitats and (2) Phorids, by modifying *A. instabilis* behavior, indirectly affect coffee berry borer abilities to invade coffee berries. Further, we examined two potential mechanisms that could be driving differences in NCE of phorids on ants in the two habitat types. Specifically, we examined whether (1) phorid density differs in complex versus simple shade habitats, and (2) sunlight has an effect on phorid ability to detect and attack *A. instabilis* ants. If, for example, habitat complexity enhances trait-modification of ants by phorids, then we would expect the trait-mediated indirect effects of phorids on coffee berry borer to be stronger in complex habitats, and coffee berry borer outbreaks to be worse. However, if trait-modification of ants by phorids were stronger in simple habitats, we would expect coffee berry borer outbreaks to be worse in simple habitats.

Materials and Methods

Study Site. We conducted our research on a large, 280 ha coffee farm in Chiapas, Mexico, during the wet season of June–July 2009. We conducted field studies in five sites within Finca Irlanda, an organic coffee farm located at 15° 11' N, 90° 20' W between 950 and 1,150 m (Philpott et al. 2008). Three of the study sites were located in low-shade areas of the farm managed with an average of 28% canopy cover. Two of the study sites were located in a high-shade area of the farm with an average of 91% canopy cover. The management in the low-shade sites most closely approximates a mix of commercial polyculture and shade monoculture whereas the high-shade sites are best described as traditional polyculture (Moguel and Toledo 1999, Philpott et al. 2008). The five sites each comprised 10–13 sampled trees (see below) and each site was separated by a minimum of 100 m. To confirm management differences in the different sites, we also recorded the percent shade surrounding all *A. instabilis* colonies used in the observations described below. We used a convex spherical densitometer to measure the amount of shade directly over the nest, and 5 m away from the tree in each cardinal direction.

Phorid-Ant Interactions and Habitat Complexity. To study whether NCE of phorid flies on ants differ between simple and complex habitats, we observed the influences of phorid flies on ant activity in both high-shade and low-shade sites within the coffee farm. We located a total of 32 independent *A. instabilis* nests in the low-shade sites ($n = 10$, $n = 12$, $n = 10$) and 22 independent *A. instabilis* nests in the high-shade sites ($n = 12$, $n = 10$) all located on *Alchornea latifolia* Klotzsch trees, a common shade tree throughout the farm. To determine whether phorid flies had stronger effects on *A. instabilis* ants in one habitat type or the other, we placed five tuna baits (5 g each) on tree trunks between 1.0–1.5 m above ground level. Then we counted the number of ants at each bait every 2 min from 2 to 30 min after placing baits (Philpott et al. 2004). At each tree, we also recorded (1) presence or

absence of phorids during the 30 min trial, and (2) time elapsed to first phorid arrival to the baits. We did not record the number of phorid individuals, or the time intervals where phorids were specifically present. We conducted observations on 21 d between 4 June and 11 July 2009. We determined statistical differences between the number of ants on trees with and without phorids using a linear mixed model with SPSS 16.0. In the model, we included number of ants as the dependent variable, phorid presence, habitat type (high-shade or low-shade), and time as fixed factors and tree nested within site and habitat type as a random factor. We also included time as a repeated factor to test for temporal autocorrelation in the errors. We ran a full factorial design for all interactions of phorid, shade type, and time, and used a first order, autoregressive covariance matrix (AR1). We also examined parameter estimates to examine for potential differences among time steps in different shade types and with or without phorids. Data for ant numbers were square root transformed to meet conditions of normality.

We examined differences in phorid density in the two habitat types as one possible mechanism for any observed difference in phorid influences on ant behavior in each habitat. We first measured the phorid density in each habitat, defined as the number of phorid attacks on worker ants during 5 min (Vandermeer et al. 2008). Phorid density is commonly assessed by examining the number of attacks, rather than by collecting individuals because of the disturbance aspirating phorids at ant nests causes (Vandermeer et al. 2008, Philpott et al. 2009). We collected ≈ 60 worker ants from a colony and placed them in two plastic sandwich tubs painted with insect-a-slip (BioQuip Products, Rancho Dominguez, CA) so that the ants could not escape. We placed the lids on the tubs and carried them to another *A. instabilis* colony located >10 m away from the tree where the ants were collected. We did this to ensure that the ant colony was not disturbed before the start of the observation. Adjacent to the colony, we hung the two tubs on stakes ≈ 1 m above ground and 0 m from the base of the tree. At the start of the observation, we uncovered the tubs, smashed three ants to release chemical pheromones that attract phorids (Mathis et al. 2011), and recorded (1) the time elapsed until phorid first arrived at the tub and (2) the number of phorid attacks on the ants within 5 min (Philpott et al. 2009). Phorid attacks were defined as individual oviposition attempts by phorids on an *A. instabilis* worker. We examined numbers of attacks on *A. instabilis* in tubs around 46 nests in the low-shade sites and 10 nests in the high-shade sites.

To examine another possible mechanism for any observed differences in phorid effects on ants in different habitats, we observed whether presence of sun flecks reaching the ground affected phorid attacks near ant nests. When *A. instabilis* are alarmed, they emit chemical cues that attract phorids from longer distances; near to the colony *Pseudacteon* sp. phorids rely on visual cues to locate *A. instabilis* hosts (Mathis

et al. 2011). It is specifically because of an inability to see ant hosts that phorids are not active at night (Feener and Brown 1993, Wuellner et al. 2002). To examine whether the light intensity at the ground level was a mechanism resulting in differences in attack rates between high-shade and low-shade habitats, we observed phorid attacks in both high light intensity sun flecks and in low light intensity patches (e.g., in shadows) in both habitat types. We collected ants and placed them in tubs for observations as described above. We located eight trees in the low-shade sites and 11 trees in the high-shade sites where the ground around the base of each tree was partly surrounded with sun flecks (high light intensity) and partly surrounded with shadows (low light intensity). We hung the two tubs on stakes ≈ 1 m above ground and at 0 m from the base of the tree. We then recorded the phorid arrival time and number of phorid attacks within 5 min. We used a paired *t*-test to compare phorid arrival time and number of phorid attacks between high light intensity and low light intensity points in each habitat type.

Phorid-Ant-Coffee Berry Borer Interactions. To examine the cascading effects of phorid flies on coffee berry borer predation by ants, we conducted lab experiments in insect arenas (BioQuip Products). The insect arenas were $60 \times 60 \times 60$ cm tents with mesh and clear plastic walls and floors. In each arena, we placed a coffee branch with 10 undamaged berries and between 4 and 9 leaves, and then assigned arenas to one of three treatments: (1) with 20 coffee berry borers, (2) with 20 coffee berry borers and 20 *A. instabilis* workers, and (3) with 20 coffee berry borers, 20 *A. instabilis* workers, and two female phorid flies. We put drops of honey on the leaves of all branches to mimic scale insects commonly tended by *A. instabilis* in the field. After 24 h, we counted the number of fruits with coffee berry borer damage. We had 26 replicates of each treatment. We compared the number of coffee berry borers entering fruits in the three treatments with analysis of variance (ANOVA) and determined specific differences among treatments with Tukey's post hoc tests.

To assess whether phorids may indirectly impact coffee berry borer in the field, we examined correlations between number of phorid attacks (oviposition attempts) and coffee berry borer attack of coffee fruits around *A. instabilis* colonies. We used the data for phorid attacks to assess phorid density at each colony as outlined above. At those same 46 colonies in low-shade and 10 colonies in high-shade sites, we measured the abundance of coffee berry borers on the four coffee bushes nearest to each *A. instabilis* nest by counting the number of coffee berries per plant, up to 100, that were infected with coffee berry borer. We selected berries systematically sampling from the top of the coffee plant toward the base. We then used simple linear regressions to determine the relationship between number of phorid attacks and coffee berry borer abundance at each nest in the field.

Although at the time observations were conducted we believed there was only one species of phorid at

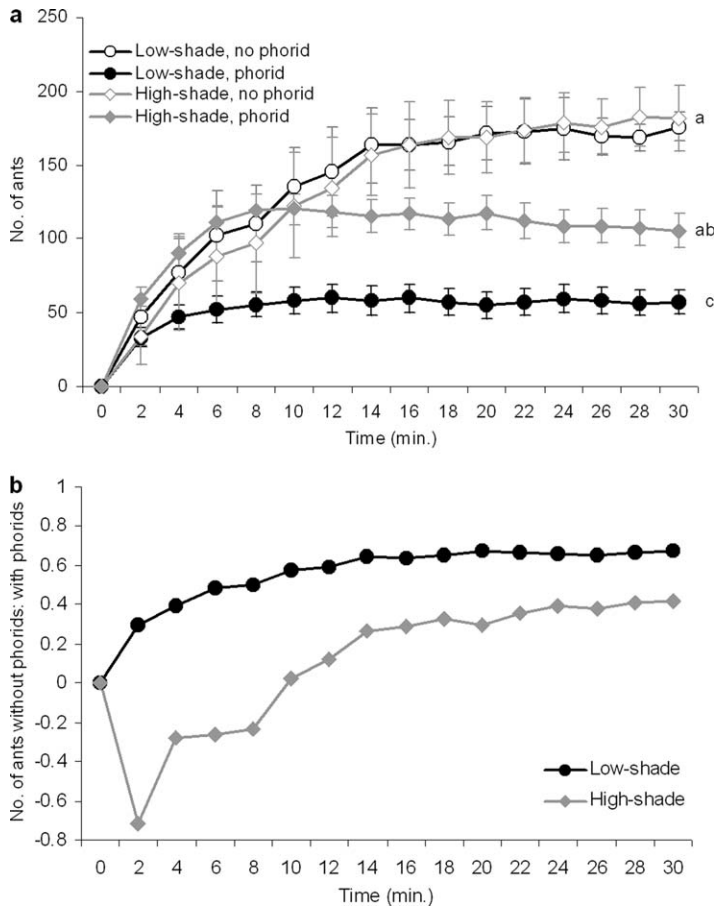


Fig. 1. Ant recruitment with and without phorid flies in the low-shade habitat and high-shade habitat. (a) Number of *A. instabilis* ants recruiting to tuna baits on *Alchornea latifolia* shade trees over 30 min in the presence (open symbols) and absence (closed symbols) of phorid flies. Error bars represent SE, and letters denote significant differences. (b) Ratio of number of ants without phorids to number of ants with phorids at tuna baits in the high-shade and low-shade habitats. Phorids decreased ant recruitment by 54% in the low-shade habitat and by 11% in the high-shade habitat.

our study site, we have since been altered that there are four morphospecies (S. Porter, B. Brown, personal communication) of phorids known to attack *A. instabilis*. At the time the research was conducted, we did not have this knowledge, and therefore did not differentiate between phorid species. Thus, the observations conducted in the field, and the lab experiments with field-collected phorids report the effects of an unknown mix of the two phorid species on the behavior of *A. instabilis*. Voucher specimens of both morphospecies of phorid flies were deposited at the Natural History Museum of Los Angeles County and also at El Colegio de la Frontera Sur, Tapachula, Chiapas, Mexico.

Results

Over the 30-min period, phorid flies significantly reduced ant foraging activity in both the low-shade and high-shade coffee habitats ($F = 10.701$, $df = 1$, 51.35 , $P = 0.002$; Fig. 1a). The number of ants recruit-

ing to the baits did not differ in high- and low-shade habitats ($F = 2.828$, $df = 1$, 42.47 , $P = 0.10$); however, there was a significant phorid by shade interaction ($F = 4.787$, $df = 1$, 48.63 , $P = 0.034$). Phorid flies limited *A. instabilis* activity more than twice as much in the low-shade habitat than in the high-shade habitat (Fig. 1b). The numbers of ants on baits increased with time ($F = 20.548$, $df = 14$, 576.71 , $P < 0.001$), and there was a significant interaction of phorid by time ($F = 5.059$, $df = 14$, 576.71 , $P < 0.001$). The difference between numbers of ants on trees with and without phorids became significant after 12 min ($t < -2.851$, $P < 0.005$). The number of ants recruiting in different habitats was not influenced by time ($F = 0.569$, $df = 14$, 576.71 , $P = 0.89$) and there was no significant three-way interaction of phorid by shade by time ($F = 0.383$, $df = 14$, 576.71 , $P = 0.98$). Values recorded at different time steps were highly autocorrelated (ARI ρ , Wald $Z = 54.149$, $P < 0.001$), but there was no significant effect of the nesting of shade tree within

Table 1. Time elapsed to first phorid attack and total no. of phorid attacks (mean ± SE) on *A. instabilis* ants during 5 min trial periods in patches of high light intensity (e.g., sun flecks) and low light intensity (shadows) around shade trees in high-shade and low-shade coffee habitats

Habitat	Light intensity	Time to first phorid attack (s)	No. of phorid attacks
High-shade	Low	165.46 ± 34.71	14.13 ± 4.22
High-shade	High	129.8 ± 33.20	22.53 ± 7.74
Low-shade	Low	132.75 ± 49.31	15.75 ± 5.73
Low-shade	High	109.5 ± 39.89	22 ± 6.41

site and shade area (ARI ρ , Wald $Z = -1.087$, $P = 0.277$).

We examined two potential mechanisms for differences in phorid effects in high- and low-shade habitats. First, we examined differences in phorid attacks in the two habitat types as a measure of phorid density. There was no significant difference between number of phorid attacks in 5 min in the high-shade (18.35 ± 5.34) and low-shade (21.43 ± 2.92) sites ($t = 2.00$, $df = 54$, $P = 0.727$). Second, we did not find any significant difference in phorid arrival or attacks in either habitat depending on light availability. The number of phorid attacks in 5 min did not differ depending on light intensity (shadows vs. sun flecks) in either the high-shade habitat (Table 1; $t = -1.129$, $df = 14$, $P = 0.139$) or in the low-shade habitat (Table 1; $t = -0.773$, $df = 7$, $P = 0.232$). Although phorids tended to arrive later in the low light intensity patches (shadows) around trees than in the high light intensity patches (sun flecks), there was no significant difference in either habitat (Table 1; high-shade habitat, $t = 1.04$, $df = 14$, $P = 0.157$; low-shade habitat, $t = -0.368$, $df = 7$, $P = 0.361$).

Phorids had significant effects on the ability of *A. instabilis* to deter coffee berry borer attack on coffee fruits. In the lab experiment, nearly twice as many fruits were attacked by coffee berry borer in arenas without *A. instabilis* and with both *A. instabilis* and phorids than in arenas with only coffee berry borer and *A. instabilis* ($F = 4.326$, $df = 2, 67$, $P = 0.017$; Fig. 2).

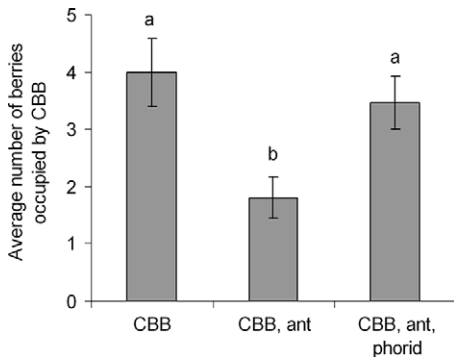


Fig. 2. Coffee berry borer (CBB) infestation level in treatments: coffee berry borer alone, with *A. instabilis*, and with *Pseudacteon* spp. phorid flies and *A. instabilis*. Error bars represent SE, and letters denote significant differences.

According to Tukey’s post hoc tests, a significantly lower fraction of fruits were attacked by coffee berry borer in the ant treatment than in the coffee berry borer-only arenas ($P = 0.021$) or the coffee berry borer-ant-phorid arenas ($P = 0.046$), but the coffee berry borer-only and coffee berry borer-ant-phorid treatments did not differ ($P = 0.868$).

In the field, however, we did not observe any relationship between phorid attacks and fruits attacked by the coffee berry borer. The number of phorid attacks in 5 min was not significantly correlated with the proportion of attacked fruits on coffee plants near *A. instabilis* colonies ($y = 0.0002x + 0.0195$, $R^2 = 0.0124$, $P = 0.531$).

Discussion

Pseudacteon sp. phorids had greater NCE on *A. instabilis* ants in a simple habitat than a complex habitat. Thus, behavioral modifications of ants by *Pseudacteon* parasitoids may be affected by changes in habitat, and may have important implications not only for ecological interactions involving *A. instabilis* ants but also for the red imported fire ant (*Solenopsis invicta* Buren) that is also attacked by several species of *Pseudacteon* (Wuellner and Saunders 2003). Previous research has examined ant-phorid interactions in different habitats for two other genera of parasitoids. First, *Myrmoscarius* and *Neodohrniphora* spp. phorid flies attack *Atta cephalotes* L. and *Atta sexdens* L. less on the forest edge than in the forest interior in Brazil (Almeida et al. 2008). Further, *Apocephalus* sp. phorid flies of *P. diversipilosa* and *P. bicarinata* attack significantly less in areas that contained more leaf litter because ants could escape from attacks by hiding under leaves (Wilkinson and Feener 2007). These results are similar to our experiment demonstrating that phorid flies had less of an effect on *A. instabilis* in areas with high-shade, however, because *A. instabilis* are arboreal-nesting ants, the potential mechanism driving this interaction must differ.

There are several mechanisms that may drive stronger NCE of *Pseudacteon* sp. on *A. instabilis* in low-shade compared with high-shade habitats. The two possible mechanisms we tested to explain this difference (phorid density and light availability) were not significant for our data set. Phorid density is commonly measured by counting the number of attacks because of the difficulty in assessing the density of ant-attacking phorids (e.g., Philpott et al. 2009). Specifically, in our data set, we used the number of phorid attacks during 5 min as a surrogate for phorid density. However, in ant recruitment trials, most phorids did not attack *A. instabilis* in the high-shade habitat until after 8 min. Thus, longer observation periods may be necessary to find differences between phorid densities in the two habitats. We were unable to measure phorid density again during the field season when experiments were conducted, but this is something that could be examined in the future. Second, we examined whether phorid attack rates in the two habitats differed in high light intensity patches (sun flecks) and

low light intensity patches (shadows). Phorids locate ants with visual cues, and such cues may be more difficult to perceive under low light availability. However, we did not find any differences in phorid attacks between high and low light intensity patches in either coffee habitat. It is possible that even high light intensity sun flecks in the high-shade habitat were not as bright as low-light intensity patches in the low-shade habitat. Thus, future studies could include more detailed measurements of light intensity (e.g., Leaf Area Index; Chen and Cihlar 1996) or could use experimental manipulation of light intensity to more carefully examine phorid attack differences.

Several additional factors may have influenced observed differences in phorid effects on *A. instabilis* in the two habitat types. First, wind could disturb ants causing them to emit alarm pheromones that attract phorids. Wind could also spread the chemical pheromones further in the farm attracting phorids from further away. Previous evidence shows that wind speed is generally lower in high-shade coffee habitats (Beer et al. 1998), and thus *A. instabilis* may have been more frequently disturbed in the low-shade habitat leading to higher attack rates. At our study site, however, the high-shade habitat is in a steeper valley that may have more wind than the low-shade habitat. Weather could have also impacted the activity level of *A. instabilis*. During periods of rain, ants such as the *S. invicta*, reduce foraging activity by 40% (Porter and Tschinkel 1987). Foraging behavior is reduced because rain may momentarily block exit holes, and wash away pheromones (Porter and Tschinkel 1987). Although we sampled ant-phorid interactions under the same weather conditions and at the same time each day, other weather factors, such as rain earlier during the night or early AM may have affected ant activity at observation points. Temperature also affects the maximum number of attacking adult phorids (Wuellner and Saunders 2003), and temperatures in the low-shade habitat are greater (Lin 2008), perhaps leading to greater attack rates in the low-shade habitat type. Thus, it is possible that other differences between the habitat types may affect the phorid attacks and effects on *A. instabilis* ants. Finally, two experts have viewed phorids from the study site and confirmed that there are at least four different species of phorids that attack *A. instabilis* (S. Porter, B. Brown, personal communication). Preliminary data collected since conducting this study indicates that a larger phorid morphospecies generally attacks more frequently in both habitats (K. Mathis, unpublished data). However, because two species of *Pseudacteon* attack *A. instabilis* at the study site, differences in NCE in the two habitat types may result from different densities or activity levels of multiple phorid species, a research area that is currently underway.

In the lab, presence of phorid flies cancelled out the negative impacts of *A. instabilis* on coffee berry borers, demonstrating that the phorids have a positive, trait-mediated indirect effect on coffee berry borer. This is the first concrete evidence that *A. instabilis* are important biological control agents of coffee berry borer,

the most economically important pest of coffee (Soto-Pinto et al. 2002). In the field, however, we did not find a significant relationship between number of phorid attacks and the percentage of berries infested by the coffee berry borer. The lack of a TMII between the phorid flies and the coffee berry borers in the field may be because phorids have indirect positive effects on other predators of the coffee berry borer. For example, when *A. instabilis* foraging activity is lowered by phorids, other ant species gain more access to food resources (Philpott 2005). Thus other predator species may have compensated for the reduced impacts of *A. instabilis* in areas with high phorid abundance. Some other ants, such as twig-nesting ants that live in hollow branches on coffee bushes also have significant negative impacts on coffee berry borer. In lab trials, *Pseudomyrmex ejectus* F. Smith, *P. simplex* F. Smith, *P. PSW-53*, and *Procryptocerus hylaesus* Kempf prey on borers and prevent them from infesting coffee fruits (Larsen and Philpott 2010). Further, many insectivorous birds prey on coffee berry borers (Kellermann et al. 2008), and some insectivorous birds spend less time foraging on trees with *A. instabilis* nests (Philpott et al. 2005). Where phorid flies reduce *A. instabilis* activity, bird activity and bird predation on coffee berry borers may increase. Other predators such as bats, lizards, or spiders may also prey on coffee berry borers, and the potential exists that activity of these other taxonomic groups may increase where *A. instabilis* activity is suppressed by phorids. A future experiment could be performed to determine the multi-predator effects on coffee berry borers in the presence and absence of phorids.

Phorid flies negatively affect *A. instabilis* by limiting their foraging activity, and may limit the impacts of *A. instabilis* as biological control agents. Although many may worry that phorids may be detrimental to biological control, in this case, the phorid flies actually contribute to an autonomous system of biological control involving complex interactions between ants, scale insects, coccinellid beetles, the coffee berry borer, fungal diseases, and parasitic wasps of the scales and coccinellids (Vandermeer et al. 2010). In farms with complex shade, pest control services are enhanced by higher biodiversity and higher abundance of important predators (Perfecto et al. 2004, Van Bael et al. 2008). This study demonstrates that *A. instabilis* is an important biological control agent of the coffee berry borer, and that its effects may be hindered by phorid flies. These effects of *A. instabilis* may be more important in high-shade farms, given changes in ant-phorid interactions with habitat type.

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References Cited

- Abrams, P. A. 1995. Implications of dynamically variable traits for identifying, classifying and measuring direct and indirect effects in ecological communities. *Am. Nat.* 146: 112–134.
- Abrams, P. A. 2007. Defining and measuring the impact of dynamic traits on interspecific competition. *Ecology* 88: 2555–2562.
- Almeida, de Walkiria, R., R. Wirth, and I. L. Leal. 2008. Edge-mediated reduction of phorid parasitism on leaf-cutting ants in a Brazilian Atlantic forest. *Entomol. Exp. Appl.* 129: 251–257.
- Beer, J. R. Muschler, D. Kass, and E. Somarriba. 1998. Shade management in coffee and cacao plantations. *Agroforest. Syst.* 38: 139–164.
- Chen, J. M., and J. Cihlar. 1996. Retrieving Leaf Area Index in boreal forests using Landsat TM images. *Remote Sens. Environ.* 55: 153–162.
- Consoi, F. L., C. T. Wuellner, S. B. Vinson, and L. Gilbert. 2001. Immature development of *Pseudacteon tricuspis* (Diptera: Phoridae), an endoparasitoid of the red imported fire ant (Hymenoptera: Formicidae). *Ann. Entomol. Soc. Am.* 94: 97–109.
- Feener, D. H., and B. V. Brown. 1993. Oviposition behavior of an ant-parasitizing fly, *Neodohnniphora curvinervis* (Diptera, Phoridae) and defense behavior by its leaf-cutting ant host *Atta cephalotes* (Hymenoptera, Formicidae). *J. Insect Behav.* 6: 675–688.
- Feener, D. H. 2000. Is the assembly of ant communities mediated by parasitoids? *Oikos* 90: 79–88.
- Folgarait, P. J., and L. E. Gilbert. 1999. Phorid parasitoids affect foraging activity of *Solenopsis richteri* under different availability of food in Argentina. *Ecol. Entomol.* 24: 163–173.
- Grabowski, J. H. 2004. Habitat complexity disrupts predator-prey interactions but not the trophic cascade on oyster reefs. *Ecology* 85: 995–1004.
- Grabowski, J. H., and D. L. Kimbro. 2005. Predator-avoidance behavior extends trophic cascades to refuge habitats. *Ecology* 86: 1312–1319.
- Kellermann, J. L., M. D. Johnson, A. M. Stercho, and S. C. Hackett. 2008. Ecological and economic services provided by birds on Jamaican Blue Mountain coffee farms. *Conserv. Biol.* 22: 1177–1185.
- Larsen, A., and S. M. Philpott. 2010. Twig-nesting ants: the hidden predators of the coffee berry borer. *Biotropica* 42: 342–347.
- Liere, H., and A. Larsen. 2010. Cascading trait-mediation: disruption of a trait-mediated mutualism by parasite-induced behavioral changes. *Oikos* 119: 1394–1400.
- Lin, B. B. 2008. Microclimate effects on flowering success in coffee agroforestry systems. *American-Eurasian J. Agric. Environ. Sci.* 3: 148–152.
- Marino, P. C., and D. A. Landis. 1996. Effect of landscape structure on parasitoid diversity and parasitism in agroecosystems. *Ecol. Appl.* 61: 276–284.
- Mathis, K. A., S. M. Philpott, and R. Moreira. 2011. Parasite lost: Chemical and visual cues used by *Pseudacteon* in search of *Azteca instabilis*. *J. Insect Behav.* 24: 186–199.
- Menalled, F. D., P. C. Marino, S. H. Gage, and D. A. Landis. 1999. Does agricultural landscape structure affect parasitism and parasitoid diversity? *Ecol. Appl.* 9: 634.
- Moguel, P., and V. M. Toledo. 1999. Biodiversity conservation in traditional coffee systems of Mexico. *Conserv. Biol.* 13: 11–21.
- Müller, C. B., and H. C. Godfray. 1999. Indirect interactions in aphid-parasitoid communities. *Res. Popul. Ecol.* 41: 93–106.
- Perfecto, I., R. A. Rice, R. Greenberg, and M. E. Van der Voort. 1996. Shade coffee: a disappearing refuge for biodiversity. *BioScience* 46: 598–608.
- Perfecto, I., J. H. Vandermeer, G. L. Bautista, G. I. NunPez, R. Greenberg, P. Bichier, and S. Langridge. 2004. Greater predation in shaded coffee farms: the role of resident neotropical birds. *Ecology* 85: 2677–2681.
- Perfecto, I., and J. Vandermeer. 2006. The effect of an ant-Hemipteran mutualism on the coffee berry borer (*Hypothenemus hampei*) in southern Mexico. *Agric. Ecosyst. Environ.* 117: 218–221.
- Philpott, S. M., J. Maldonado, J. Vandermeer, and I. Perfecto. 2004. Taking trophic cascades up a level: behaviorally-modified effects of phorid flies on ants and ant prey in coffee agroecosystems. *Oikos* 105: 141–147.
- Philpott, S. M. 2005. Trait-mediated effects of parasitic phorid flies (Diptera: Phoridae) on ant (Hymenoptera: Formicidae) competition and resource access in coffee agroecosystems. *Environ. Entomol.* 34: 1089–1094.
- Philpott, S. M., R. Greenberg, and P. Bichier. 2005. The influence of ants on the foraging behavior of birds in an agroforest. *Biotropica* 37: 467–470.
- Philpott, S. M., W. J. Arendt, I. Armbrrecht, P. Bichier, T. V. Diestch, C. Gordon, R. Greenberg, I. Perfecto, R. Reynoso-Santos, L. Soto-Pinto, et al. 2008. Biodiversity loss in Latin American coffee landscapes: reviewing evidence on ants, birds, and trees. *Conserv. Biol.* 22: 1093–1105.
- Philpott, S. M., I. Perfecto, J. Vandermeer, and S. Uno. 2009. Spatial scale and density dependence in a host parasitoid system: an arboreal ant, *Azteca instabilis* and its *Pseudacteon* phorid parasitoid. *Environ. Entomol.* 38: 790–796.
- Polis, G. A., A.L.W. Sears, G. R. Huxel, D. R. Strong, and J. Maron. 2000. When is a trophic cascade a trophic cascade? *Trends Ecol. Evol.* 15: 473–475.
- Porter, S. D., and W. R. Tschinkel. 1987. Foraging in *Solenopsis invicta* (Hymenoptera: Formicidae): effects of weather and season. *Environ. Entomol.* 16: 802–807.
- Schmitz, O., V. Krivan, and O. Ovadia. 2004. Trophic cascades: the primacy of trait-mediated indirect interactions. *Ecol. Lett.* 7: 153–163.
- Sih, A., G. Enlund, and D. Wooster. 1998. Emergent impacts of multiple predators on prey. *Trends Ecol. Evol.* 13: 350–355.
- Soto-Pinto, L., I. Perfecto, and J. Caballero-Nieto. 2002. Shade over coffee: its effects on berry borer, leaf rust and spontaneous herbs in Chiapas, Mexico. *Agroforestry Syst.* 55: 37–45.
- Swisher, B. J., D. A. Soluk, and D. H. Wahl. 1998. Non-additive predation in littoral habitats: influences of habitat complexity. *Oikos* 81: 30–37.
- Trussell, G. C., E. J. Ewanchuck, and M. D. Bertness. 2002. Field evidence for trait-mediated indirect interactions in a rocky intertidal food web. *Ecol. Lett.* 5: 241–245.
- Van Bael, S. A., S. M. Philpott, R. Greenberg, P. Bichier, N. A. Barber, K. A. Mooney, and D. S. Gruner. 2008. Birds as predators in tropical agroforestry systems. *Ecology* 89: 928–934.
- Vandermeer, J., I. Perfecto, and S. M. Philpott. 2008. Clusters of ant colonies and robust criticality in a tropical agroecosystem. *Nature* 451: 457–460.
- Vandermeer, J., I. Perfecto, and S. M. Philpott. 2010. Ecological complexity and pest control in organic coffee

- production: uncovering an autonomous ecosystem service. *Bioscience* 60: 56–526.
- Werner, E. E., and S. D. Peacor. 2003. A review of trait-mediated indirect interactions in ecological communities. *Ecology* 84: 1083–1100.
- Wilkinson, E. B., and D. H. Feener. 2007. Habitat complexity modifies ant–parasitoid interactions: implications for community dynamics and the role of disturbance. *Oecologia* 152: 151–161.
- Wuellner, C. T., Dall'Aglio-Holvorcem, W. W. Benson, and L. E. Gilbert. 2002. Phorid fly (Diptera: Phoridae) oviposition behavior and fire ant (Hymenoptera: Formicidae) reaction to attack differ according to phorid species. *Ann. Entomol. Soc. Am.* 95: 257–266.
- Wuellner, C. T., and J. B. Saunders. 2003. Circadian and circannual patterns of activity and territory shifts: comparing a native ant (*Solenopsis geminata*, Hymenoptera: Formicidae) with its exotic, invasive congener (*S. invicta*) and its parasitoids (*Pseudacteon* spp., Diptera: Phoridae) at a Central Texas Site. *Ann. Entomol. Soc. Am.* 96: 5.

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